Remarks

Claims 58-87 were pending in the subject application. Applicants gratefully acknowledge the Examiner's withdrawal of the rejection under 35 USC §101. Accordingly, claims 58-87 are currently before the Examiner for consideration. Favorable consideration of the pending claims is respectfully requested.

Claim 72 is objected to on the grounds that it does not further limit the scope of claim 58 from which it depends. Applicants respectfully assert that claim 72 does limit the scope of claim 58. Claim 72 can be understood to refer to "direct" labelling as opposed to indirect labelling where labelling is via the intermediary of a binding pair such as streptavidin-biotin, as described in the subject specification (*e.g.*, see page 20 of the specification). The direct labelling of claim 72 refers to a link with the nucleotide that is, for example, covalent. That claim 72 recites that there is direct labelling does not mean that the label is not attached to the nucleotide via a cleavable linker. Thus, Applicants respectfully assert that claim 72 does further limit claim 58. Accordingly, reconsideration and withdrawal of the objection to claim 72 is respectfully requested.

The drawings have been objected to as not complying with the drawing requirements. Submitted herewith are replacement drawings for Figures 4 and 6-8. Figures 6, 7, and 8 have been amended by increasing the font size of the chemical structures. In addition, Figures 4, 7, and 8 are now labeled as Figures 4A, 4B, 7A, 7B, 8A, and 8B. Entry and consideration of the replacement drawings is respectfully requested. Accordingly, reconsideration and withdrawal of the objection is respectfully requested.

Claims 58-87 are rejected under 35 USC §112, first paragraph, as non-enabled by the subject specification. The Examiner asserts that the claimed sequencing method is not enabled for the full scope of the claims. Applicants respectfully assert that the claimed invention is enabled by the subject specification. In order to meet the enablement requirement under 35 USC §112, first paragraph, a specification must describe how to make and how to use the claimed invention such that a person of ordinary skill in the relevant art would be able to practice the invention without resort to undue experimentation. *In re Glass*, 181 USPQ 31 (CCPA 1974). Applicants also respectfully submit that a significant amount of experimentation can be permitted under the enablement

requirement. *Ex parte Jackson*, 217 USPQ 804, 807 (Bd. Pat. App. & Int. 1982) ("The test [for undue experimentation] is not merely quantitative, since a considerable amount of experimentation is permissible, if it is merely routine . . .") (emphasis added).

In section 9 of the Office Action, the Examiner asserts under the §112 rejection that the specification fails to teach the sequencing of any nucleic acid "where the label is retained." Applicants respectfully assert that in the claimed method, the label is not retained on the nucleotide and is in fact cleaved from it (as is recited in step (c) of claim 58). Thus, the claimed invention is not directed to a method of sequencing a nucleic acid where the label is retained on the incorporated nucleotide and, therefore, the specification need not teach nucleic acid sequencing where the label is retained. However, if the Examiner's intended point of contention is that unreacted labelled nucleotide is retained in the same solution after incorporation and that the cleaved label is also retained in the same solution after cleavage and the Examiner is suggesting these could thereby interfere with detection of the incorporated nucleotide, then Applicants respectfully request that the Examiner clarify the grounds for rejection. Applicants maintain, in this regard, that the claimed invention can be practiced by an ordinarily skilled artisan. The subject specification teaches a number of embodiments in which the method can be practiced wherein unreacted labelled nucleotide and cleaved label do not interfere. These include, for example, embodiments wherein a washing step is performed after each of steps **a-c** of the claimed invention. Thus, the unreacted labelled nucleotide and the cleaved label would be washed away. In other embodiments, the labelled nucleotide can comprise a quencher moiety. This quencher moiety is intended to quench the label on any unreacted nucleotide. In this embodiment, the sequencing process can be performed while labelled (but quenched) unreacted nucleotides are present in the solution. The unreacted labelled nucleotide does not fluoresce and, therefore, would not interfere with detection of the label on the reacted nucleotide. After reaction, the nucleotide incorporated into the polynucleotide would be devoid of the quencher and so its fluorescence would then be detectable. Therefore, having the unreacted labelled nucleotide in the same solution (usually in excess) would not pose a problem in practicing the claimed method.

Applicants also respectfully assert that the subject specification describes means for practicing the claimed method in regard to the cleaved label. First, the cleaved label would not be in

excess and, secondly, it would be able to diffuse away from the location (the surface) where the reaction is taking place, into the three-dimensional space available around the reaction location. Therefore, due to the small amount of cleaved label produced and because it would diffuse away from the reaction and detection location, the presence of cleaved label in the same solution would not present a problem. Moreover, the specification also teaches how detection can be carried out using Total Internal Reflection Fluorescence (TIRF) Microscopy (see, for example, second paragraph, page 62 of the specification) in which illumination is from an exponentially decaying evanescent wave, which means that as the label diffuses away from the reaction location it will become less and less bright and, therefore, background would be further minimized. The subject specification also describes imaging by FRET, which means only labels in the very close vicinity (a few nm) of an appropriately labelled reaction location would be detected and the majority of the label in solution would not be detected. The subject specification also teaches, at page 21, paragraph 2, that a combination of surfaces with low adsorption of fluorescent nucleotides with quencher nucleotide may be especially advantageous. This provides a means to prevent unreacted or cleaved label from sticking to the surface, which would further prevent the unreacted labelled nucleotides and cleaved labels from causing problems in detection of the incorporated nucleotide. In addition, page 28, paragraph 4, of the specification describes treatment with or inclusion of blockers which prevent non-specific surface binding.

Applicants further note that, at the time of the subject invention, assays that used quenchers were available (Taqman), that used FRET (Big-Dye sequencing) were available, washing after carrying out reaction steps on a surface (*e.g.*, microarray experiments) was known, surfaces with low surface adsorption of labelled nucleotides (*e.g.*, Braslavsky *et al.* 2003) were known, and methods that limited penetration of illumination (TIRF and Zero-mode waveguides) were known. Accordingly, Applicants respectfully assert that there is sufficient detail and disclosure in the application to enable a person of ordinary skill in the art to carry out the invention without recourse to unduc experimentation.

In regard to the Examiner's comments under section 10 of the Office Action, Applicants respectfully assert that an application for patent does not have to teach actual working examples and/or devices as long as a person of ordinary skill in the art, having the benefit of the teachings of

the specification, would be able to practice the claimed method. *In re Borkowski*, 164 USPQ 642 (CCPA 1970). Applicants respectfully submit that the co-inventor's publication (Mir *et al.*, *Nucleic Acids Research*, January 2009, 37(1):e5) (copy attached) describes how three (3) contiguous bases of sequencing information are obtained using one of the detection schemes (microarray scanner) described in the subject specification using equipment available in a typical lab but without specific instrumentation for temperature or fluid control. The fluidics was all done manually in the work reported in the Mir *et al.* publication and did not require a specific new device. Applicants submit that a fluidics device is not absolutely necessary to carry out the sequencing on a "multitude of nucleic acids in a simultaneous manner"; rather, it would just make the work less labor intensive if the aim was to go through a high number of iterations of steps **a-c** in independent claim 58. Moreover, detection devices that can be used with the claimed method are commercially available (*e.g.*, microarray scanner, microscope). The "use" of a microscope or microarray scanner does not involve any further knowledge than a person of ordinary skill in the relevant art would already possess.

With regard to the Examiner's specific point that no "temperature control, or fluid communication and fluid control" have been enabled, Applicants respectfully assert that such special measures are not necessary for practicing the claimed method. In some embodiments, all the reaction and wash steps can be carried out manually (as in Mir *et al.*, *supra*) and do not need fluid communication and fluid control. Automation of the fluidics would be an option but is not a requirement for practicing the claimed invention. In other embodiments (see, for example, claim 59), fluid communication and fluid control would not be needed as the reaction would be homogeneous. With regard to temperature control, no specific device out of the ordinary is needed. A typical molecular biology lab has several means of temperature control at their disposal, be it temperature controlled ovens, water baths, heating blocks or PCR machines. A microarray or molecular cytogenetics lab would typically have PCR or other programmable temperature varying devices that enable temperature control of samples. Thus, suitable temperature and fluid control means are known in the art and a person of ordinary skill in the art would be able to practice the claimed method without undue experimentation.

In Section 11 of the Office Action, the Examiner asserts that the specification teaches (at page 11, lines 1-2) that sequencing will involve the use of extraordinary long linkers but that the claims do not require linkers of any particular length. Applicants respectfully assert that the use of linkers as discussed at page 11 of the subject specification applies to specific embodiments of sequencing methods, and in particular those methods where the label is not cleaved from the nucleotide after incorporation. In Applicants' claimed invention, the label is cleaved from the incorporated nucleotide. Therefore, Applicants respectfully assert that the section of the specification referred by the Examiner is not of particular relevance to the claimed invention, and its enablement.

In section 12 of the Office Action, the Examiner asserts that numerous labels are recited but that the specification has not set forth reaction conditions under which each of the specified/claimed reagents are to be used in the claimed method. Applicants respectfully assert that conditions for reactions utilizing nucleotides labelled with relevant moieties (*e.g.*, Fluorescein, Texas Red, Cy3 and Cy5 (cyanine)) are provided in the subject specification. For example, the third paragraph on page 73 of the specification describes using a thermostable polymerase. Although reaction conditions for every single dye or dye group set forth in claims 61, 62, and 67 are not provided, appropriate conditions would be readily apparent to the ordinarily skilled artisan and can be extended to the other dyes listed in the claims and are applicable regardless of whether the moiety functions as a fluorescence emitter or as a quencher. Applicants note that page 24, first paragraph, of the subject specification describes one specific emitter-quencher pair as fluorescein and Dabcyl. Another specific nucleotide that is described on page 24 of the subject specification is Pyrrolo-dCTP-dabcyl which is stated in the specification to be incorporated by thermostable polymerases (examples of reaction conditions are provided on page 73 of the specification). At the time of the subject invention, and as stated in the specification, Internally Quenched Nucleotide fluorescent reporters (such as Pyrrolo-dCTP-dabcyl) were described in PCT publication WO 03/089670 (referenced in the subject specification) and are commercially available for real-time PCR, microarray technologies, and diagnostics from Lawler Scientific/Glen Research (Sterling, VA). The person of ordinary skill in the art would have been able to prepare and/or purchase these reagents and use them (in the claimed methods) according to conditions known in the art and/or the vendor recommended reaction

conditions, without undue experimentation. It is well settled in patent law that a specification is not required to teach that which is well known in the art. *Hybritech v. Monoclonal Antibodies, Inc.*, 231 USPQ 81 (Fed. Cir. 1986). Furthermore, as the Examiner is aware, every aspect of a generic claim need not have been carried out by an inventor, or exemplified in the specification, and only a reasonable amount of detail must be provided in order to enable members of the public to understand and carry out the invention. *Brenner v. Manson*, 148 USPQ 689 (1966). Applicants respectfully assert that the subject specification provides sufficient disclosure and sufficient detail for an ordinarily skilled artisan to make and use the claimed method.

In view of the above remarks, reconsideration and withdrawal of the rejection under 35 USC §112, first paragraph, is respectfully requested.

In view of the foregoing remarks, Applicants believe that the currently pending claims are in condition for allowance, and such action is respectfully requested.

The Commissioner is hereby authorized to charge any fees under 37 CFR §§1.16 or 1.17 as required by this paper to Deposit Account 19-0065.

Applicants invite the Examiner to call the undersigned if clarification is needed on any of this response, or if the Examiner believes a telephonic interview would expedite the prosecution of the subject application to completion.

Respectfully submitted,

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Attachments: Figures 1-11 (13 total sheets)
Mir *et al.* publication